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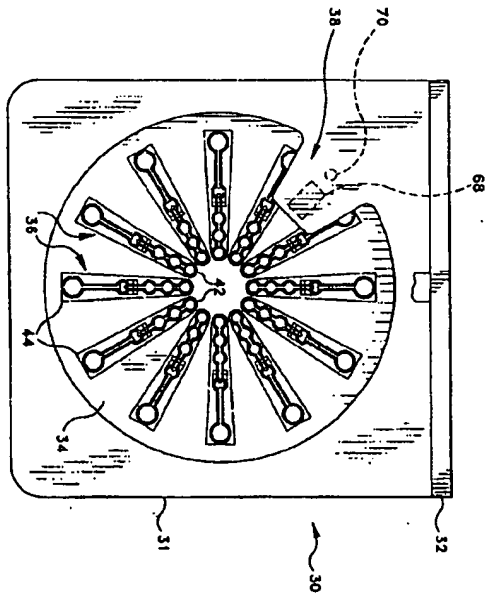
(54) Method and apparatus for fully automated nucleic acid amplification, nucleic acid assay and immunoassay

(57) A disposable, self-contained test unit is provided for use in carrying out an immunoassay or an integrated nucleic acid amplification and nucleic acid assay. The test unit includes sample, reagent and waste chambers, and the flow of sample and reagent liquids is controlled by centrifugal force. Different liquids may be caused to flow at different times by locating the chambers at different radial distances from the axis of rotation, and progressively increasing the speed of rotation during the assay procedure. An automated test instrument for receiving and rotating a number of such test units is also disclosed.

FIG-2



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Description

Cross-Reference to Related Application

Related subject matter is disclosed and claimed in a co-pending U.S. patent application of Hugh V. Cottingham, Serial No. 08/213,304, filed on March 14, 1994 and entitled "Nucleic Acid Amplification Method and Apparatus", the disclosure of which is expressly incorporated herein by reference, and also the corresponding EP 95102870.3.

Field of the Invention

The present invention relates to methods and apparatus for performing automated clinical diagnostic assays, particularly immunoassays or integrated nucleic acid amplifications and nucleic acid assays, and to a self-contained, disposable test unit for use in such methods and apparatus.

Background of the Invention

The advent of nucleic acid amplification techniques offers great potential to improve the sensitivity of detection in many areas. One such area is in the detection and identification of human pathogens. The ability to detect as little as one organism makes detection possible at a much earlier stage in the progression of a disease. Unfortunately, while nucleic acid amplification techniques are extremely powerful, they are not simple to perform. Complications with respect to operating parameters, such as precise time-temperature requirements, and possible contamination by amplified nucleic acids have so far restricted these techniques to the research laboratory. In order to make these amplification techniques practical for use in hospitals and clinical laboratories, problems arising from complicated operating parameters and contamination must be resolved. Moreover, for reasons of manufacturing efficiency, and due to the emphasis on reduction of health care costs, it is also desirable to provide a universal platform for nucleic acid amplification, nucleic acid assay, immunoassay and assays using nucleic acid ligands to bind proteins or small molecular weight molecules (hereinafter referred to as nucleic acid ligand based assays). Such nucleic acid ligand based assays may also utilize bidirectional nucleic acid ligand compounds.

One way to meet these objectives is to construct a fully automated system to run the entire nucleic acid amplification and nucleic acid assay (or immunoassay), convert the raw data into clinical results, and report the results through the local area network (LAN) of a laboratory or hospital. In order to make such a system practical, all of the steps involved in the test procedure should preferably be carried out while the liquid biological sample remains confined within a single, disposable test unit. This not only reduces the risk of contamination of the laboratory or hospital environment, but also allows the test procedure to be carried out quickly, inexpensively and without the need for highly skilled personnel.

Unfortunately, existing designs for self-contained test units are not entirely satisfactory, particularly when cost factors are considered. For example, U.S. Patent No. 5,229,297, to Schnipelsky et al, describes a cuvette for DNA amplification and detection which comprises a plurality of flexible compartments for containing a sample, amplifying reagents and detection reagents, together with passageways connecting the sample and reagent compartments with a detection site and a waste compartment. A roller is used to squeeze or compress the sample and reagent compartments in a desired sequence, thereby forcing the sample and detection reagents through the passageways to the detection site and waste compartment. Temporary seals are used to isolate the sample and reagent compartments from the passageways until sufficient pressure is generated by the roller. Although the disclosed arrangement is advantageous in that the sample remains within the cuvette during amplification and detection, the need for a roller to break the temporary seals and cause the various fluids to flow between compartments makes it difficult to automate the procedure, particularly when a number of samples and cuvettes are to be processed at the same time. In an alternative embodiment of the Schnipelsky et al patent, the fluids are moved between compartments by movable pistons which form a part of the cuvette itself. Again, however, automated processing of several cuvettes at once is difficult with this embodiment, and the need for several pistons in each cuvette renders the cost of the cuvette higher than might be desired.

It is therefore an object of the present invention to provide a fully automated system to perform an integrated nucleic acid amplification and nucleic acid assay, or to perform an immunoassay or nucleic acid ligand based assay.

It is another object of the invention to contain all reagents and components necessary to perform immunoassays or nucleic acid ligand based assays, or integrated nucleic acid amplifications and nucleic acid assays, for a particular sample in a single, disposable test unit.

It is a further object of the invention to provide a disposable test unit which can be automatically closed and permanently sealed to prevent contamination of the laboratory environment from amplified nucleic acids.

It is a still further object of the invention to support nucleic acid amplification multiplexing and report a multiplicity of nucleic acid assay results using a single, disposable test unit and a single clinical sample.

It is yet another object of the invention to support multiplexed immunoassays and report a multiplicity of immunoassay results using a single, disposable test unit and a single clinical sample.

It is yet another object of the invention to support multiplexed nucleic acid ligand based assays and report